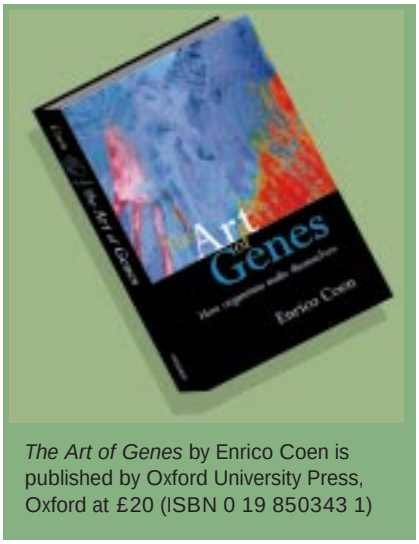
bindweed to make them twist round their supports in a right-handed helix. It contains a wealth of interesting material, and Coen communicates his immense learning with a hundred appealing tales.

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## Quick guide

# Molten globules

Christina Redfield

***What are they?*** Molten globules are partially folded proteins that have some characteristics of both folded (or 'native') and unfolded proteins. Like a native protein, the classic molten globule is compact and has native secondary structure, such as α-helices and β-sheets; like an unfolded protein, it lacks specific native tertiary interactions.

***When were they discovered?*** As early as the 1960s, there was evidence that under certain conditions protein molecules might have properties intermediate between the rigid native and fully unfolded states. The concept of such a protein as an intermediate in protein folding was suggested by the late Oleg Ptitsyn in the 1970s. A compact, partly folded protein was first observed experimentally in the early 1980s and the term molten globule was coined in 1983.

***How can I make one?*** Take a protein and dissolve it under mildly denaturing conditions, such as low pH, low concentrations of urea, elevated temperature or a lack of cofactors like metal ions or heme groups. But beware, only certain proteins form molten globules, the best-studied being β-lactoglobulin, carbonic anhydrase, apomyoglobin, cytochrome *c* and α-lactalbumin.

***How do I know if I have one?*** A variety of biophysical techniques, including circular dichroism, can be used to detect the hallmarks of a molten globule: lack of specific fixed tertiary contacts and the presence of native secondary structure.

***What do they look like?*** Who knows? The structural analysis of molten globules is difficult because they are flexible structures and consist of an interconverting ensemble of conformations, so it's hard to apply standard X-ray and NMR techniques. Advances in NMR, however, mean that some structural information is beginning to emerge for apomyoglobin and α-lactalbumin.

***Do they exist*** in vivo***?*** Possibly. Transient structures detected in studies of the folding of certain small proteins are similar to the stable molten globules that can be generated under mildly denaturing conditions. So, whether or not molten globules represent genuine *in vivo* folding intermediates, they could provide insights into the nature of transient folding intermediates that are, well, too transient to study in detail.

***When is a molten globule not a molten globule?*** The term molten globule has been used to describe a wide range of partly folded states of proteins, giving rise to a lively debate as to which of these conform to the original definition. The term 'pre-molten globule' has been used for an intermediate between the unfolded protein and the molten globule. Confusingly, some authors have also used the term pre-molten globule to describe a more structured state of the α-lactalbumin molten globule. The term 'highly-ordered molten globule' has been used to describe an intermediate between the molten globule and the native protein. Because they have stable tertiary contacts, it has been possible to determine the structure of the highly-ordered molten globules of apocytochrome $b_{562}$ and interleukin-4 using NMR. But their tertiary contacts also mean that these systems might be better described as native proteins with regions of significant dynamic disorder. Will the real molten globules please stand up?

***Do they have any biological significance?*** The misfolding and aggregation of proteins is now known to be involved in several diseases, including cystic fibrosis and the amyloid diseases. A protein that fails to fold properly and gets trapped in a folding intermediate — such as the molten globule — could be mislocated in the cell, or aggregate and be converted to a non-native fibrillar form.

***Where can I find out more?***

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